Immunotoxicology Committee
Cytokine release assay ring trial
Ellen Evans, PhD, Pfizer

Bioaccumulation Committee
Fish hepatic metabolism ring trial
Kelly Faye, PhD, EPA
ITC: Cytokine Release Assay Ring Trial

Ellen W. Evans, DVM PhD DACVP, Pfizer
Case Study Overview

• How did we get here?

• Why is this important to address now?

• What is the project to address the issues?
How did we get here?

- TGN 1412 (CD28 superagonist) cytokine storm in phase 1
- EMA workshop
- HESI ITC Survey and Workshop

What did the ITC learn?

- No “gold standard”

- Multiple platforms in multiple locations

- Decisions based on MOAs and other needs

- Within an approach: Different conditions (e.g. standards, dilutions, etc.)
How did we get here?

2006
TGN 1412 (CD28 superagonist) cytokine storm in phase 1

2009
EMA workshop

2013
HESI ITC Survey and Workshop

2016
Launch of HESI ITC – NIBSC standards study
The National Institute for Biological Standards and Control

- Global leader in the characterisation, standardisation and control of biological medicines.
- UK’s Official Medicines Control Laboratory for biological medicines
- World leader in development and production of international standards for biologics
- NIBSC plays a major role in assuring the quality of biological medicines worldwide through the provision of biological reference materials, by testing products and carrying out research.
Why now?

• Qualification & validation of platforms
• Future context
  – Comparison of results between platforms
  – New biotherapeutics
  – Harmonization of existing assays to reduce improper immunotox evaluation
HESI ITC – NIBSC Standards Study

• Institutions already have optimized in-house protocols
• **NOT** looking to change current methodologies

**BUT RATHER HAVE A SET OF STANDARDS THAT...**

- Fit into current assays already in use
- Have a response range (low, moderate, high)

• Standard repository at NIBSC for future availability
Example of assay formats

**mAb in Solid Phase (SP)**

- Purified PBMC
- Whole blood
- 10% diluted blood

**mAb in Aqueous Phase (AQ)**

- Purified PBMC
- Whole blood
- 10% diluted blood

2 day precultured PBMC

How well do these standards perform across platforms, and in comparison to in-house controls

Stebbings et al J. Immunol 2007; 179:3325-3331
What standards will be evaluated?

- **Positive Controls**
  - Anti-human CD52 mAb; Human IgG1 (Mild cytokine release)
  - Anti-human CD3 mAb; mouse IgG2a (Moderate cytokine release)
  - Anti-human CD28 mAb: human IgG4 (severe cytokine release)
- **Negative Controls, non-specific human anti-4-hydroxy-3-nitrophenyl acetyl (NP) isotypes**
  - Human anti-NP; IgG1,
  - Mouse anti-NP; IgG2a,
  - Human anti-NP; IgG4
- **10x PBS (Gibco, ref 70013-016, lot 1697466)**
HESI ITC – NIBSC Standards Study

GOAL: Data gathered June – September 2016 with publication in 2017

Controls:
3 positive
3 negative
Summary

**Hazard** – cytokine storm

**Efficient** – quick study and incorporation into existing methods

**Standards** – incorporating a set of standards to allow for harmonization and future evaluation

**Integrity** – repository for future use and data confidence across platforms